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FEEDING DETERRENCE OF ANTHRAQUINONE, ANTHRACENE, AND ANTHRONE TO RICE-EATING BIRDS

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Abstract: Safe, effective bird repellents are needed as seed treatments and for many other agricultural uses. Quinones are distributed widely in nature and many have predator defense and antiherbivory functions. One compound, 9,10-anthraquinone, was identified as a bird repellent in the 1940s, but is not registered for use in the United States. We evaluated it and 2 structurally related compounds, anthrone and anthracene, for repellency to rice-eating birds. In choice tests with individually caged red-winged blackbirds (*Agelaius phoeniceus*) anthraquinone and anthrone produced comparable reductions in consumption of treated rice at rates of 0.05, 0.10, and 0.25% (g/g). At 0.50%, however, only anthraquinone suppressed consumption of untreated rice as well as treated rice. Anthracene was least effective of the 3 compounds and was tested only at 0.50%. In 1-cup tests, consumption of anthraquinone-treated rice by individual blackbirds was suppressed at 0.10, 0.25, and 0.50%. Rice consumption by individually caged female boat-tailed grackles (*Quiscalus major*) exposed to the 0.50% treatment was similar to that of redwings at the 0.10% treatment. In choice tests of 3-bird groups in large flight enclosures, red-winged blackbirds discriminated strongly against 0.25% anthraqunione-treated rice. Observations of videotaped birds revealed no evidence of contact irritation or unpleasant taste; rather post-ingestive illness, as evidenced by one vomiting bird, suggests that anthraquinone repellency is due to learned behavior.

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Key words: *Agelaius phoeniceus*, anthraquinone, anthrone, bird repellent, boat-tailed grackle, crop protection, feeding deterrent, *Quiscalus major*, red-winged blackbird, rice, seed treatment.

Quinones are distributed widely among a number of plant and invertebrate animal taxa (Thomson 1987). Benzoquinones and derivative compounds used for predator defense have been identified in secretions of numerous arthropods (Eisner and Meinwald 1966).

Anthraquinone compounds make up the largest group of natural quinones (Sherburne 1972). Most are found in plants, and although their functions are not well-documented, 1 compound, emodin (1-3-8-trihydroxy-6-methyl-anthraquinone; Fig. 1) is a potent antifeedant (Sherburne 1972). Anthraquinones also occur in many invertebrates and some appear to have a predator defense function (Hilker and Köpf 1994).

In wildlife management, one compound, 9,10-anthraquinone (hereafter, anthraquinone; Fig. 1), for many years has been recognized as an avian feeding deterrent. The first United States patent for this use was obtained in 1944 (Heckmanns and Meisenheimer 1944). In the United States, evaluation of anthraquinone as a bird repellent has emphasized protection of pine (*Pinus* spp.) and rice seeds (Mann et al. 1956, Royall and Neff 1961). In extensive evaluation of potential treatments of rice seed, Neff and Meanley (1957) considered anthraquinone the standard against which candidate bird-repellent chemicals were compared; however, others found anthraquinone less effective. Reportedly, pigeons (*Columba livia*) are insensitive to aqueous solutions of anthraquinone (Duncan 1963), and rooks (*Corvus frugilegus*) are not deterred from newly sown corn treated with 5 or 10% (by mass) anthraquinone (Wright 1962). Although not registered as a bird repellent in the United States, anthraquinone is used in Europe.

The current lack of effective, registered bird-repellent chemicals for seed treatment makes anthraquinone an interesting candidate, particularly because of its demonstrated effectiveness as a rice seed treatment at rates of 1.0% (g/g) and higher (Neff and Meanley 1957). In addition, anthraquinone was an effective feeding deterrent to red-winged blackbirds, house sparrows (*Passer domesticus*), and yellow-headed blackbirds (*Xanthocephalus xanthocephalus*) at seed treatment rates of 0.10–0.60% (g/g) with no alternate food available (Schafer et al. 1983). Our study was conducted to elucidate further

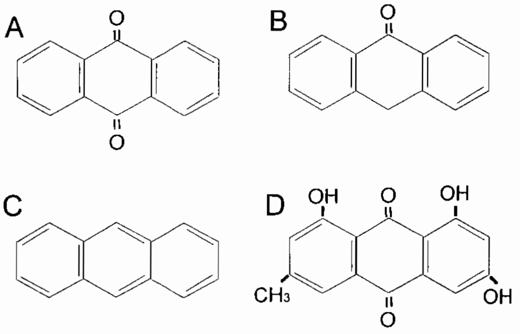

Fig. 1. Chemical structures of anthraquinone (A), anthrone (B), anthracene (C), and emodin (D).

the bird repellency of anthraquinone and 2 related compounds, anthrone and anthracene (Fig. 1), to the red-winged blackbird and the boat-tailed grackle. Each spring, these species cause substantial damage to newly planted rice in the southeastern United States (Wilson et al. 1989, Decker et al. 1990), and effective nonlethal means to reduce such losses currently are lacking.

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METHODS

We obtained 9,10-anthraquinone (Chem. Abstracts Serv. Registry No. 84-65-1), anthracene (120-12-7), and anthrone (90-44-8) from Aldrich Chemical Company, Milwaukee, Wisconsin. Purity of each chemical was listed as 97%. We treated rice seed 3–5 days before the initial test day by mixing the appropriate amount of chemical with 25 mL of propylene glycol (trial 1) or with a commercial adhesive (trials 2–4) and then applied the mixture to 1 kg of rice seed in a rotating tumbler. Treated seed was airdried for 2 hours and then stored in airtight containers in an air-conditioned lab until used. No chemical analyses were performed.

We captured birds in decoy traps in Alachua County, Florida, and housed them by species in communal cages (1.2 × 1.2 × 1.7 m) in a roofed outdoor aviary 2–6 months before testing. Unless otherwise stated, birds had free access to water and maintenance food, Quail Starter (Hillandale Farms, Lake Butler, Fla.). Each bird was banded and released following the study. Results are reported as means ± SE.

Trial 1, 2-cup Cage Test (0.50% Anthraquinone, Anthracene, and Anthrone)

We removed male red-winged blackbirds from holding cages, determined mass, and assigned them at random to individual test cages (45 × 45 × 45 cm) to form 3 treatment groups of 5 birds each. After 3 days of acclimation to the smaller cages, we tested birds for 3 hours on 4 consecutive mornings. We removed maintenance food at 0700 hours and presented test food at 0800 hours. Each bird received 2 cups, 1 with 20 g of untreated rice (propylene glycol only) and the other with 20 g of rice treated with 0.50% (g/g) anthraquinone, anthracene, or anthrone. We determined the position of the treated cup in each cage by a coin flip on day 1 and alternated it daily thereafter. Aluminum pans suspended beneath each cup caught spillage. We placed food cups containing each treatment in vacant cages to determine moisture gain or loss. We removed test food at 1100 hours, replaced the maintenance food, and determined consumption by subtraction after correction for spillage and changes because of moisture. After the final test day, we determined mass, banded, and released each bird.

We evaluated rice seed consumption among treatments (3), days (4), and cups (2) in a 3-way repeated measures analysis of variance (ANOVA). We used Tukey's HSD test (Steel and Torrie 1980) to isolate differences ($P < 0.05$) among means. A 1-way ANOVA was used to test for changes in body mass of birds among treatment groups. Before analysis, we applied the arcsin transformation to percent changes.

Trial 2, 2-cup Cage Test (0.05, 0.10, 0.25% Anthraquinone and Anthrone)

Testing and analysis followed the same procedures as in trial 1 except that we evaluated only anthrone and anthraquinone and we used 3 treatment levels of 5 birds each plus a 0% group that received rice with the adhesive only. We selected 1 bird in the 0.25% anthraquinone group to be videotaped so we could observe reactions to the treatment.

Trial 3, 1-cup Cage Test (0.10. 0.25, 0.50% Anthraquinone)

Testing procedures followed those in trials 1 and 2 except only 1 food cup was used. We tested male red-winged blackbirds (n = 6 birds/level) with rice seed treated at 0.10, 0.25, and 0.50% (g/g), and we also tested female boat-tailed grackles at 0.50% (n = 8 birds). There was a 3-day acclimation period followed by a 4-day pretreatment period during which the single test-food cup held plain rice. After a 2-day break, we presented rice treated with the randomly assigned level of anthraquinone for 3 hours on 4 consecutive mornings.

We analyzed results for redwings and grackles separately. For the redwings, we performed a 2-way analysis of covariance on the consumption of treated rice with anthraquinone level as the independent factor, repeated measures across days, and pretreatment consumption as the covariate. We subjected the grackle data to a 1-way ANOVA that compared consumption during the pretreatment and treatment periods. To observe reactions to the treatment, we videotaped 1 grackle each day.

Trial 4, 2-bowl Group Enclosure Test (0.25% Anthraquinone)

We placed 3 male red-winged blackbirds into each of 5 test enclosures (3.1 × 9.5 × 2.1 m) equipped with shaded perches, a waterer, and bowls at 2 covered feeding stations. During a 3-day acclimation period, each food bowl held maintenance food. By a coin flip, we assigned 1 bowl in each pen as the treatment bowl. Then, during a 4-day pretreatment phase, we presented 100 g of untreated rice daily in each of the food bowls. During the 4-day test period that followed, the treated bowl held 100 g of rice treated with 0.25% anthraquinone and the other bowl held 100 g of untreated rice. The location of the treated food bowl was alternated daily. Each bowl rested on an aluminum spillage pan. During each pretreatment and test day, we removed maintenance food at 0700 hours, and presented test food at 0800 hours. We removed test bowls and spillage pans at 1500 hours and replaced the maintenance food.

We assessed consumption in a 2-way ANOVA with bowl as the independent factor and repeated measures across days. We applied Tukey's HSD test (Steel and Torrie 1980) to isolate differences (P < 0.05) among means.

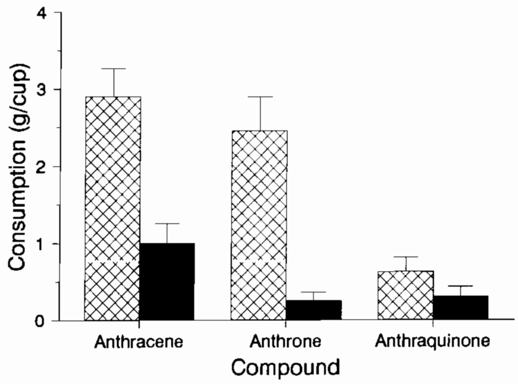

Fig. 2. Mean daily consumption by individually caged male red-winged blackbirds of untreated rice (cross-hatched) and rice seed treated with 0.5% (g/g) anthraquinone, anthrone, or anthracene (solid bars). Each bird received 2 cups of seed for 3 hours on 4 consecutive mornings. Capped bars indicate 1 SE.

RESULTS

Trial 1, 2-cup Cage Test (0.50% Anthraquinone, Anthracene, and Anthrone)

Total rice consumption varied (F = 13.35; 2, 12 df; P = 0.001) among groups, with the anthraquinone group averaging least (0.46 ± 0.11 g/cup) and the anthracene averaging most (1.97 ± 0.25 g/cup). Across all groups, consumption from the treated cup (0.52 ± 0.10 g/bird) was less (F = 51.98; 1, 12 df; P < 0.001) than that from the control cup (2.00 ± 0.23 g/bird). Consumption also varied with day (F = 5.98; 3, 36 df; P = 0.002), as birds ate less on days 1 (1.00 ± 0.20 g/cup) and 3 (1.04 ± 0.23 g/cup) than on days 2 (1.46 ± 0.32 g/cup) and 4 (1.55 ± 0.35 g/cup).

The interaction between treatment group and cup (F = 8.18; 2, 12 df; P = 0.006) reflects strongly suppressed consumption from the control cup by the anthraquinone group compared to the other groups (Fig. 2). The group × day interaction (F = 3.07; 6, 36 df; P = 0.016) indicates that whereas consumption in the anthracene group tended to increase from day 1 to day 4, consumption by the anthraquinone group remained suppressed. The strong interaction between cup and day (F = 21.86; 3, 36 df; P < 0.001) indicates decreasing consumption across days from the treated cup, which was the opposite trend in consumption from the control cup. The 3-way interaction (F = 2.85; 6, 36 df; P = 0.023) is indicative of suppressed

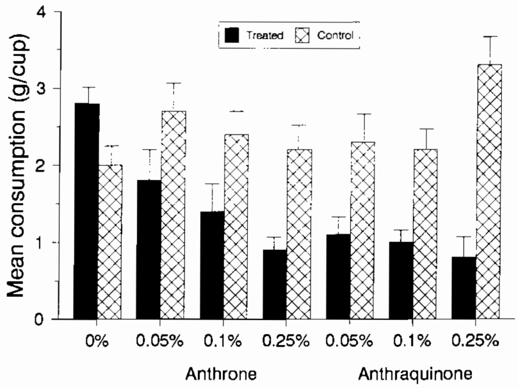

Fig. 3. Mean daily consumption by individually caged male red-winged blackbirds of untreated rice (cross-hatched) and rice seed treated with anthraquinone or anthrone (solid bars) at the rate indicated. Each bird received 2 cups of seed for 3 hours on 4 consecutive mornings. Capped bars denote 1 SE.

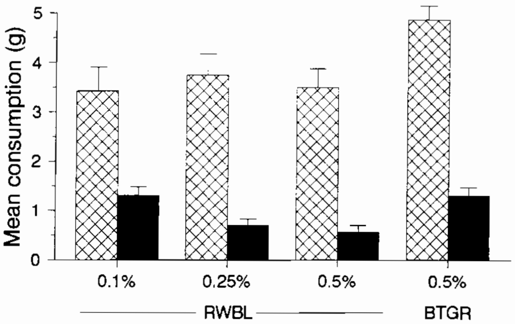

Fig. 4. Mean daily consumption by individually caged male red-winged blackbirds (RWBL) and female boat-tailed grackles (BTGR) given 1 cup of rice seed for 3 hours during pretreatment and treatment periods. Rice was untreated (cross-hatched) on each of 4 consecutive mornings during pretreatment and treated with anthraquinone (solid bars) at the rate indicated during the 4-day treatment period. Capped bars denote 1 SE.

consumption from both cups by the anthraquinone group throughout the test.

Body mass changes did not differ (P = 0.817) among groups as test birds gained mass during the feeding trial. Mean increases in body mass ranged from 4.3 ± 1.8% in the anthracene group to 6.3 ± 3.1% in the anthrone group.

Trial 2, 2-cup Cage Test (0.05, 0.10, 0.25% Anthraquinone and Anthrone)

Total consumption did not differ across groups (P = 0.061), but varied with day (F = 3.50; 3, 84 df; P = 0.019). Birds ate least on day 1 (1.74 ± 0.14 g/cup), after which consumption stabilized at 1.9–2.1 g/cup. Overall, consumption from treated cups (1.41 ± 0.11 g/bird) was markedly lower (F = 30.44; 1, 28 df; P < 0.001) than from control cups (2.42 ± 0.13 g/bird).

The interaction between treatment group and cup (F = 4.06; 6, 28 df; P = 0.005) reflected greater differences in consumption between treated and control cups at the 0.25% treatment levels relative to such differences in the other groups (Fig. 3). The interaction between cup and day (F = 5.43; 3, 84 df; P = 0.002) reflected increased differences in consumption between treated and control cups on days 3 and 4 relative to days 1 and 2, as consumption from control cups increased and consumption from treatment cups decreased.

Birds generally lost body mass throughout the trial. Mean loss of body mass was greatest in the 0.10% anthraquinone group (7.4 ± 2.1%) and least in the 0.05% anthrone group (2.3 ± 1.5%).

One bird in the 0.25% group was videotaped, and it showed a strong preference for the right side of the cage. Because of camera malfunction, we did not obtain video for day 1. On day 2, the treated cup was on the left, so the bird did not contact treated seed. On day 3, however, the treated cup was on the preferred side, and the bird ate freely from the cup and consumed 53 seeds in more than 15 minutes. For the next 11 minutes, it perched quietly and ate nothing, but then vomited 4 times during the next 7 minutes. For the remainder of the 2-hour taping, the bird ate principally from the untreated cup on the left side of the cage but continued to sample treated rice as well. On day 4, the bird again ate from the right cup, which held untreated seed.

Trial 3, 1-cup Cage Test (0.10, 0.25, 0.50% Anthraquinone)

Red-winged Blackbirds.—Consumption of treated rice differed (F = 21.67; 2, 15 df; P < 0.001) among anthraquinone levels (Fig. 4); the 0.10% group ate the most (1.31 ± 0.15 g/bird) and the 0.50% group ate the least (0.56 ± 0.12 g/bird). Overall, consumption of treated rice declined steadily across test days (F = 20.25; 3, 44 df; P < 0.001) from 1.46 ± 0.15 g/bird on day 1 to 0.40 ± 0.14 g/bird on day 4. The interaction between treatment group and day (F = 3.27; 6, 44 df; P = 0.010) reflected an increase in consumption by the 0.50% group on day 4. The increased consumption was contrary to the trend in the other groups and was caused by unusually high consumption (2.49 g) by a single bird.

Body mass changes did not differ among groups ($P = 0.909$). Loss of body mass ranged from 7.8 ± 3.6% in the 0.25% anthraquinone group to 9.2 ± 2.0% in the 0.10% group.

Boat-tailed Grackles.—Rice consumption decreased ($F = 165.42$; 1, 62 df; $P < 0.001$) from 4.86 ± 0.25 g/bird during pretreatment to 1.31 ± 0.13 g/bird during the treatment period (Fig. 4). Treated rice consumption averaged 1.91 g/bird on the first treatment day and stabilized at 1.0–1.2 g/bird thereafter. Mean loss of body mass was 5.8 ± 0.8% during the feeding trial. We videotaped 1 bird on 2 consecutive days in the treatment period. During initial daily feeding bouts, the bird ate 51 treated seeds in 16 minutes on day 1 and 10 treated seeds in 4 minutes on day 2. There was no obvious indication of irritation, distaste, or malaise on either day. Two different birds were videotaped on treatment days 3 and 4, and each ate sparingly (2.29 and 1.87 g) with no sign of discomfort or irritation.

Trial 4, 2-bowl Group Enclosure Test (0.25% Anthraquinone)

Rice consumption varied ($F = 8.83$; 6, 8 df; $P < 0.001$) among days, and was lowest (4.3 ± 1.4 g/bowl) on the second treatment day. The amount eaten differed ($F = 105.11$; 1, 48 df; $P < 0.001$) between bowls, averaging 4.5 ± 0.7 g from the treated bowl and 11.8 ± 0.9 g from the untreated bowl. The interaction between day and bowl ($F = 30.33$; 6, 48 df; $P < 0.001$) reflected almost equal consumption between bowls in the pretreatment period compared to increasing differences between bowls during the treatment period (Fig. 5).

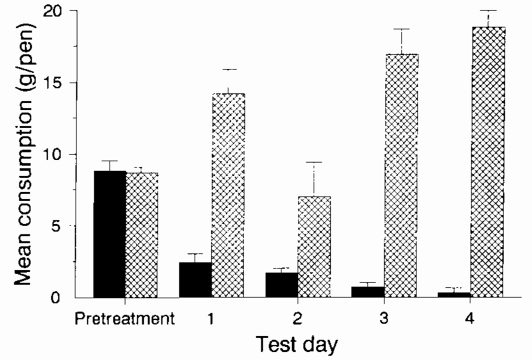

Fig. 5. Mean daily consumption by 3-bird groups of male red-winged blackbirds given 2 bowls of rice for 7 hours during 4-day pretreatment and treatment periods. During pretreatment, each bowl held 100 g of untreated rice; consumption averaged across the 4 days is shown. During the 4-day treatment period, 1 bowl in each pen held rice treated with 0.25% anthraquinone (solid bars) while the other held untreated rice (cross-hatched bars). Capped bars denote 1 SE.

DISCUSSION

Anthraquinone substantially deterred feeding by individual and group-housed red-winged blackbirds in choice tests. Furthermore, at 0.50% (g/g), the highest level tested, consumption of untreated rice was suppressed markedly as well. This result was unanticipated because such a response is unreported in feeding trials of other irritant or taste-aversion compounds, such as methyl anthranilate (Mason et al. 1991, Avery et al. 1995) or pulegone (Mason 1990, Avery et al. 1996). There was little variance in response among individual birds within the treatment group, and our finding suggests that if the anthraquinone treatment rate is sufficiently high, birds will stop eating altogether rather than attempt to distinguish untreated from treated food (Avery 1985). Alternatively, a 0.5% anthraquinone treatment might irritate the digestive tract sufficiently to suppress a bird's appetite in general. The 0.50% anthrone treatment also suppressed consumption of treated rice, but unlike birds exposed to anthraquinone, those given the anthrone treatment compensated by feeding more from the control cup (Fig. 2). At lower concentrations, there was virtually no difference between the 2 compounds (Fig. 3).

In test situations with no alternate, untreated food available, the highest anthraquinone rate tested, 0.50%, reduced consumption by red-winged blackbirds by 84% relative to pretreatment values (Fig. 4). Schafer et al. (1983) reported 50% reduction in consumption at treatment rates between 0.10 and 0.60%. For boat-tailed grackles, which are about twice as large as redwings, we recorded 71% reduction in rice consumption.

Quinones and other secondary compounds present in unripe fruit may discourage frugivores from consuming the fruit before seeds are ready to be dispersed (Sherburne 1972). Emodin (Fig. 1), an anthraquinone compound found in unripe fruit of several species of *Rhamnus*, is a cathartic that inhibits feeding by birds and mammals (Sherburne 1972). Pulp of other fruit also produces laxative effects on birds, but spe-

Table 1. Preference scores (consumption of treated rice divided by consumption of treated plus untreated rice) of individually caged red-winged blackbirds. Each bird received 2 cups of rice, and the position of the treated cup was alternated daily. Trials lasted 3 hours except for methyl cinnamate trials (6 hr). A preference score of 0.5 indicates indifference; lower scores indicate avoidance of the treatment.

Compound	Treatment rate (g/g) 0.10%	0.50%	Source
Anthraquinone	0.31	0.33[a]	This study
Anthrone	0.37	0.09	This study
Methyl anthranilate	0.46	0.32	Avery et al. 1995
Methyl cinnamate	0.47	0.45[b]	Avery and Decker 1992
Pulegone	0.13	0.06	Avery et al. 1996

[a] Total consumption (treated plus untreated) was reduced to <1 g, compared to 2.5–3.5 g total consumption in groups exposed to other compounds.
[b] Treatment rate was 0.40%, not 0.50%.

cific compounds were not identified (Murray et al. 1994).

The mode of action for anthraquinone is not certain. The compound is an irritant (Windholz 1983) and is described as bad-tasting (Thomson 1988, Schafer 1991), but there was no head-shaking or bill-wiping by video-taped red-winged blackbirds (0.25% treatment) or boat-tailed grackles (0.50%) to suggest an aversive taste. Rather, the prolonged vomiting by 1 red-wing after consuming treated rice for 15 minutes suggests that post-ingestional effects are predominant, and that birds must learn to avoid anthraquinone-treated food. This interpretation is consistent with findings of Hilker and Köpf (1994) who observed that captive tits (*Parus* spp.) readily consumed anthraquinone-treated insect larvae on the first day of experiments but reduced consumption of them thereafter, in favor of untreated larvae. Contact irritation or taste aversion possibly become operative at higher treatment rates, but there was no evidence of either at the rates we tested.

MANAGEMENT IMPLICATIONS

Our findings support continued development and evaluation of anthraquinone and anthrone as bird repellents. As seed treatments, both compounds perform better than methyl anthranilate and methyl cinnamate but appear less effective than pulegone (Table 1). Also noteworthy is the sharply reduced total consumption by birds exposed to the 0.50% anthraquinone treatment (Fig. 2). Comparative performance can be affected by numerous factors, so the information in Table 1 is restricted to male red-winged blackbirds in 2-cup tests with untreated rice as the alternate food. Additional comparative studies are warranted, including field evaluation of specific use patterns and formulations.

Technical grade anthraquinone costs $36.25/250 g (Aldrich Chem. Co., Milwaukee, Wis.). From the same source, cost of 2 other recently tested blackbird repellents was $21.80/500 g for methyl anthranilate (Avery et al. 1995) and $173.00/500 mL for pulegone (Avery et al. 1996). Because of expenses related to development and registration, the final cost of the product to the consumer inevitably differs from that of the technical material. Current information suggests that the price of an anthraquinone-based rice seed treatment will be <$30/ha (K. E. Ballinger, Jr., EBI, Wilmington, Del., unpubl. data). Because the cost to replant a heavily damaged rice field exceeds $100/ha (Holler et al. 1982), treatment of seed with anthraquinone to deter blackbirds appears to be feasible economically. Nevertheless, other important criteria, such as environmental degradation and compatability with seed germination and plant growth, have to be fully evaluated.

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